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Background: The peanut protein Ara h 1 is a major peanut allergen. The aim of our study was to compare IgE epitopes and epitope patterns for intact and digested Ara h 1 between individual rats. No previous study has compared intact and digested allergen-specific epitopes.
Methods: Brown Norway (BN) rats were immunised i.p. three times with 200 µg purified intact Ara h 1 or 200 µg gastroduodenal digestion products thereof. The digesta contained no intact Ara h 1. Ara h 1 digesta was composed of peptides ≤ 1.5 kDa of which 1/3 had aggregated to detectable complexes of M_r 4000-10000. Rat sera previously tested positive for specific antibodies in ELISA and in rat basophil leukaemia (RBL) assay were selected (Ara h 1 immunised N=4 and digesta immunised N=1). Ara h 1- as well as Ara h 1 digesta-specific IgE epitopes were assessed by competitive immunoscreening of a phage-displayed random 7-mer peptide library using polyclonal IgE from individual rat sera. The resulting peptide mimics were mapped on the surface of a model of the 3D-structure of the Ara h 1-monomer using a computer assisted epitope mapping tool (EMT).
Results: Competitive immunoscreening and epitope mapping identified rat-specific IgE epitope patterns. However, most peptide mimics were clustered into two different allergenic areas on the 3D model of the Ara h 1 monomer known to be the Ara h 1 monomer-monomer contact sites, in agreement with results obtained by Shin *et al.* 1998 using human sera. This was irrespective of whether serum was raised against intact or digested Ara h 1 and irrespective of whether competitive immunoscreening was made with intact Ara h 1 or digested Ara h 1. Likewise the physico-chemical characteristics of the amino acids in the peptide mimics were very similar, of which hydrophobic amino acids occurred most frequently. Also no significant differences were observed with respect to single amino acid distribution, amino acid fingerprinting profile, and surface accessibility of the epitope mimics.
Conclusion: Although Ara h 1 is labile to gastroduodenal digestion the resulting epitope mapping profile of the digesta showed no obvious difference from that of intact Ara h 1. This was irrespective of whether serum was raised against intact or digested Ara h 1, or whether competitive immunoscreening was made with intact or digested Ara h 1. Epitope mapping of intact and digested Ara h 1 with sera from peanut allergic patients is in progress.